Cellular transcripts regulated during infectinos with highly pathogenic H5N1 avian influenza virus in 3 host systems.

ABSTRACT

BACKGROUND: Highly pathogenic Avian Influenza (HPAI) virus is able to infect many hosts and the virus replicates in high levels in the respiratory tract inducing severe lung lesions. The pathogenesis of the disease is actually the outcome of the infection as determined by complex host-virus interactions involving the functional kinetics of large numbers of participating genes. Understanding the genes and proteins involved in host cellular responses are therefore, critical for the elucidation of the mechanisms of infection. METHODS: Differentially expressed transcripts regulated in a H5N1 infections of whole lung organ of chicken, in-vitro chick embryo lung primary cell culture (CeLu) and a continuous Madin Darby Canine Kidney cell line was undertaken. An improved mRNA differential display technique (Gene Fishing™) using annealing control primers that generates reproducible, authentic and long PCR products that are detectable on agarose gels was used for the identification of differentially expressed genes (DEGs). Seven of the genes have been selected for validation using a TaqMan® based real time quantitative PCR assay. RESULTS: Thirty seven known and unique differentially expressed genes from lungs of chickens, CeLu and MDCK cells were isolated. Among the genes isolated and identified include heat shock proteins, Cyclin D2, Prenyl (decaprenyl) diphosphate synthase, IL-8 and many other unknown genes. The quantitative real time RT-PCR assay data showed that the transcription kinetics of the selected genes were clearly altered during infection by the Highly Pathogenic Avian Influenza virus. CONCLUSION: The Gene Fishing™ technique has allowed for the first time, the isolation and identification of sequences of host cellular genes regulated during H5N1 virus infection. In this limited study, the differentially expressed genes in the three host systems were not identical, thus suggesting that their responses to the H5N1 infection may not share similar mechanisms and pathways.

Keyword: Avian influenza virus mRNA differential display; Annealing control primer; Hsp60 gene; Cyclin D2 gene; Interleukin 8 gene.